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INVESTIGATIONS ON SOME FRUIT DISEASES.

I. APPLE ROTS IN COLD STORAGE;

II. PEACH ROT IN COLD STORAGE;

III. SULPHUR FUMIGATION TO DESTROY APPLE ROT FUNGI;

IV. APPLE INJURY BY SULPHUR FUMIGATION;

V. ENLARGEMENT OF APPLE SCAB SPOTS UNDER A COVERING OF BORDEAUX MIXTURE.

H. J. EUSTACE.

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BULLETIN No. 297.

INVESTIGATIONS ON SOME FRUIT DISEASES.

H. J. EUSTACE.

SUMMARY.

Apples artificially inoculated with decay-producing fungi were placed in commercial cold storage, temperature 32° F., and held there for two months or more. Of the several species used, Penicillium glaucum, (blue mold), was the only one that developed and caused decay. Upon removal to a warmer temperature all the species of fungi developed and caused decay.

Decay was not entirely prevented in inoculated apples held in a temperature of 35° to 56°; and developed vigorously in a temperature of 48° to 69°.

Peaches inoculated with Sclerotinia fructigena (brown rot), the most common and destructive decay of peaches, developed a small amount of decay in two weeks in a temperature of 32° F.

Spores of Penicillium glaucum (blue mold), can be destroyed by fumigation with sulphur, but when these fumes come in contact with ripe apples the commercial value of the fruit is lessened.

Spraying immature apples with bordeaux mixture did not check the development of spots of Venturia inæqualis (scab), that had started previous to the application of the bordeaux mixture.

I. APPLE ROTS IN COLD STORAGE.

Since the commercial cold storage of apples has become such an important industry in New York State many questions relating to it have demanded solution. Many of these questions have been studied experimentally, and definite knowledge of great practical value regarding them determined.

However, the question as to whether the several species of fungi that most commonly cause the decay of fruit will develop and produce decay in fruit while held in commercial cold storage has never been studied.

Since disputes over the condition of fruit held in commercial cold storage houses are frequent and sometimes find their way into court, it is highly desirable to have some reliable information as to the troubles that are directly traceable to the growth of certain well known fungi.

The methods of conducting the experiments upon this question were to obtain a good supply of pure cultures of the fungi, from these to make inoculations under sterile conditions into sound apples and, to place the fruits at once in a commercial cold storage house. After remaining there a reasonable time they were removed into a warmer temperature.

Experiments were carried on in the winter of 1905 in duplicate, but in different cold storage houses. Higher temperatures were also experimented with; one from 37° to 56° F., the average being 47° F.; and another from 54° to 65.5° F., the average being 60.6° F. The work with commercial cold storage at different temperatures was repeated in the winter of 1906 in one house.

THE EXPERIMENTS OF 1905.

A good supply of vigorous pure cultures of bitter rot (*Glomerella rufomaculans* (Berk.) Sp. & vonSchr.), black rot (*Sphæropsis malorum* Pk.), blue mold (*Penicillium glaucum* Lk.), brown rot (*Sclerotinia fructigena* (Pers.) Schrt.), pink rot (*Cephalothecium roseum* Cda.) and *Alternaria* sp. were secured by taking apples that were naturally infected with

these fungi and with some of the mature spores making dilution or poured plate cultures with potato agar. From these plates transfer cultures were made to sterilized sugar beet plugs and from these several sub-cultures were made. These last cultures were then tested to make sure of their pathogenicity.

Sound mature apples of the following varieties were selected for the inoculations: Baldwin, Tompkins King, Northern Spy, Rhode Island *Greening,* Russet and Sutton. Each fruit was cleaned with a cloth and then dipped in a solution of corrosive sublimate 1: 1000; from this it was drained and rinsed in clean distilled water, drained again and the excess moisture absorbed with a cloth that had been wet with corrosive sublimate and dried.

Several fruits of each variety were inoculated with each species of fungus used.

The inoculations were made on March 9, 1905. On each fruit the epidermis was punctured in three places with a sterile knife and with another sterile knife some of the fungus from a pure culture was inserted in the puncture.

In connection with the inoculation experiments it was thought desirable to test the power of these species of fungi to grow in culture. This was done by preparing a quantity of petri dishes of sterile agar and transferring, under sterile precautions, some of the fungus from the pure cultures to several places upon the agar. The apples inoculated with a single species of fungus were put together and packed in a separate compartment in a bushel box. The plate cultures were wrapped in clean paper and packed in with the apples. Immediately after completing the work the boxes were taken by a special messenger to a commercial cold storage plant and at once placed in a room where the temperature was 31° F. Records of the temperature in this room were taken six times each day at regular intervals. The record gave 360 readings during the time the experiment was in progress; 3 were 29°, 1 was 29½°, 97 were 30°, 75 were 30½°, 143 were 31°, 37 were 31½°, and 4 were 32°.

The apples and cultures were all removed from the cold storage house on May 9 and examined that day. The results were as follows:

TABLE I.—CONDITION OF INOCULATED APPLES AND CULTURES AFTER BEING IN COLD STORAGE FOR TWO MONTHS.
FIRST TEST.

Fungus.	Growth in apples.	Growth in cultures.
Alternaria sp.	No growth in any of the fruits.	Slight growth.
Bitter rot, *Glomerella rufomaculans.*	No growth in any of the fruits.	No growth.
Black rot, *Sphæropsis malorum.*	No growth in any of the fruits.	No growth.
Blue mold, *Penicillium glaucum.*	An area of decay more than an inch in diameter has developed at every point of inoculation.	Small growth in all the petri dishes.
Brown rot, *Sclerotinia fructigena.*	No growth in any of the fruits.	No growth.
Pink rot, *Cephalothecium roseum.*	No growth in any of the fruits.	No growth.
Scab, *Venturia inæqualis.*	No experiments with fruit.	Slight growth in the dishes and on sugar beet plugs.

The condition of the apples is shown in Plate I, which is a reproduction of a photograph made the same day the fruit was removed from cold storage.

After making the notes and the photograph the apples were placed in a room where the temperature was about 70° F. during the day but somewhat less at night. Notes on the progress and development of the decay were made on May 16 and 23, the conditions being as follows:

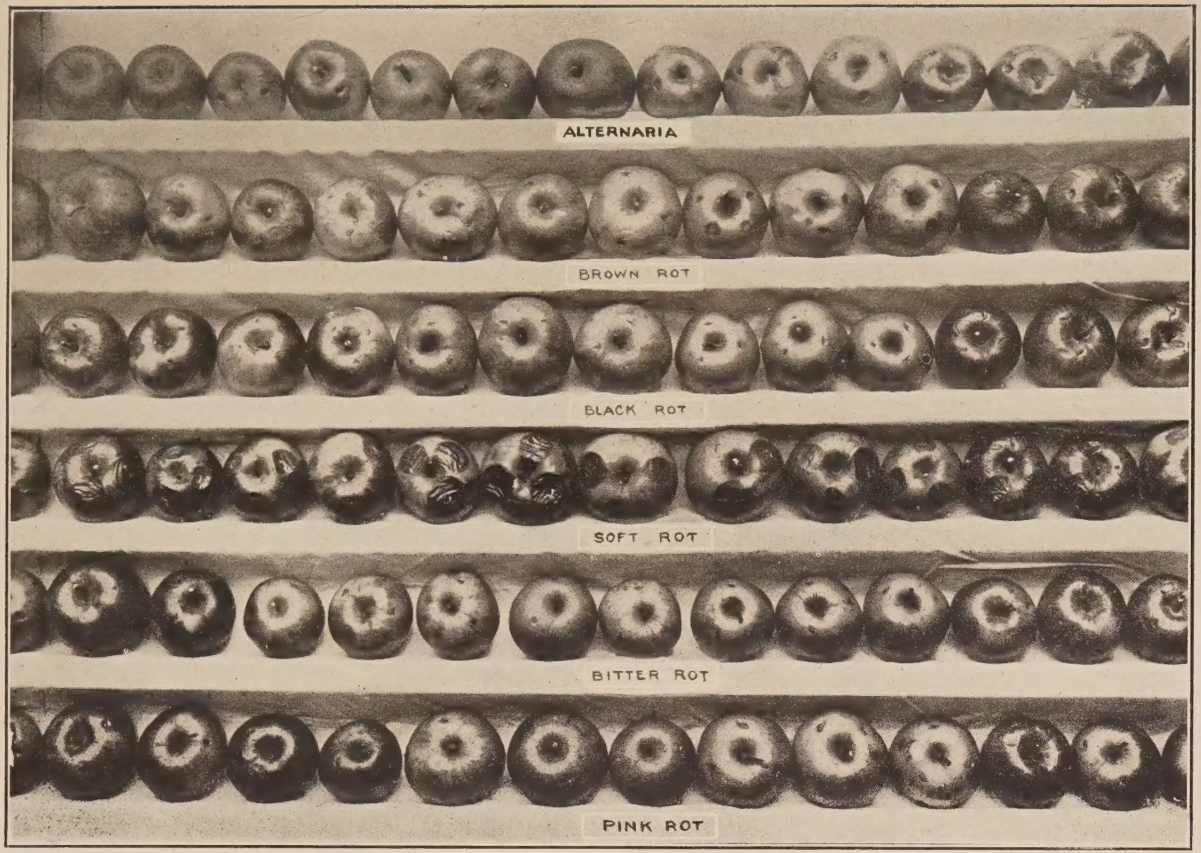

PLATE I.—CONDITION OF INOCULATED APPLES WHEN REMOVED FROM COLD STORAGE WAREHOUSE.

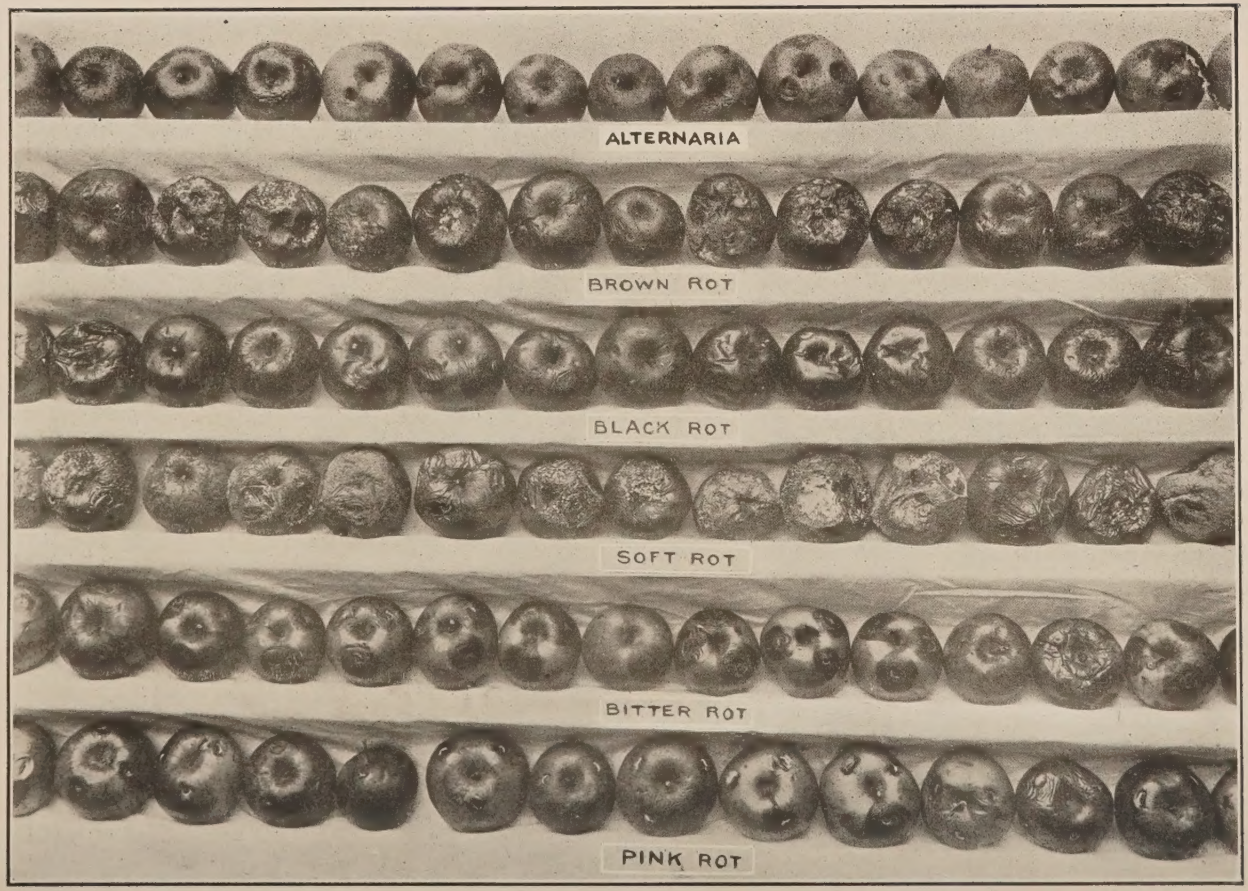

PLATE II.—CONDITION OF APPLES SHOWN IN PLATE I AFTER THEY HAD BEEN IN A WARM ROOM FOR TWO WEEKS.

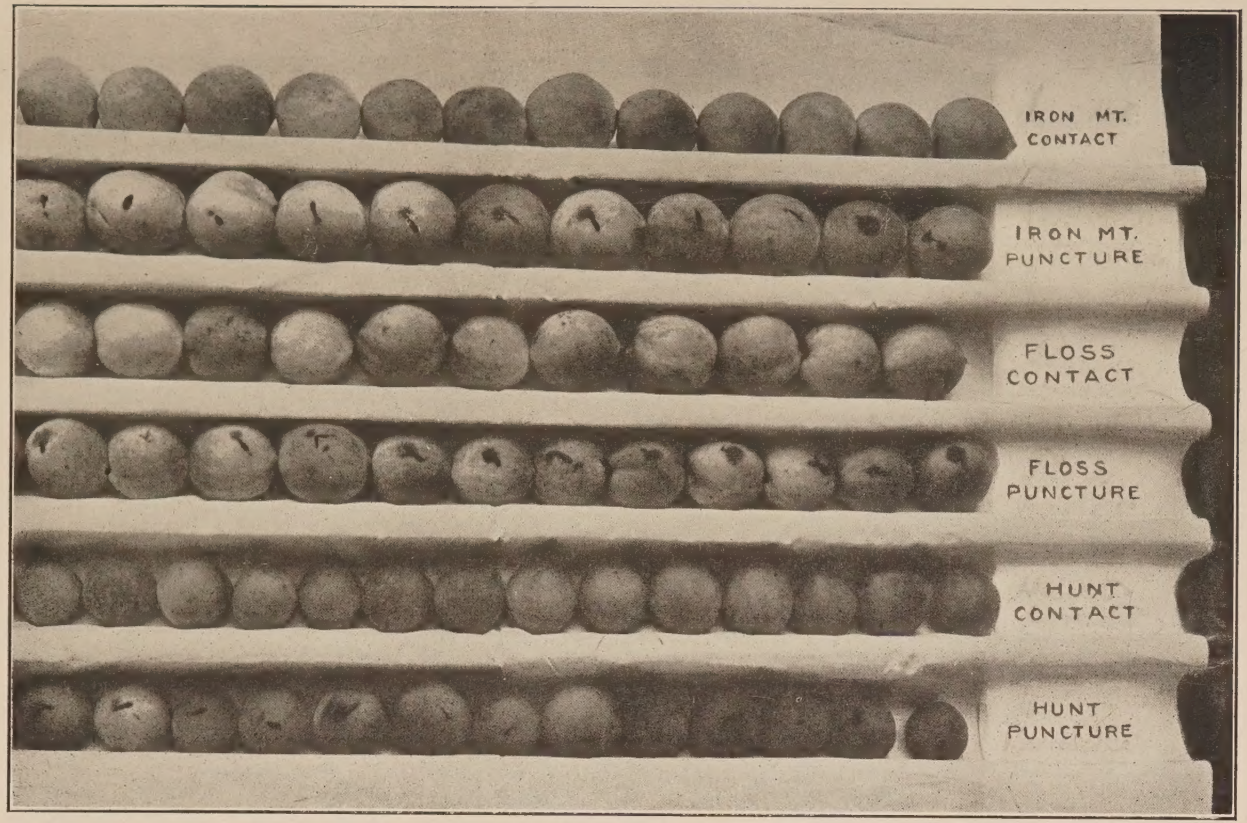

PLATE III.—CONDITION OF INOCULATED PEACHES WHEN REMOVED FROM A COLD STORAGE WAREHOUSE.

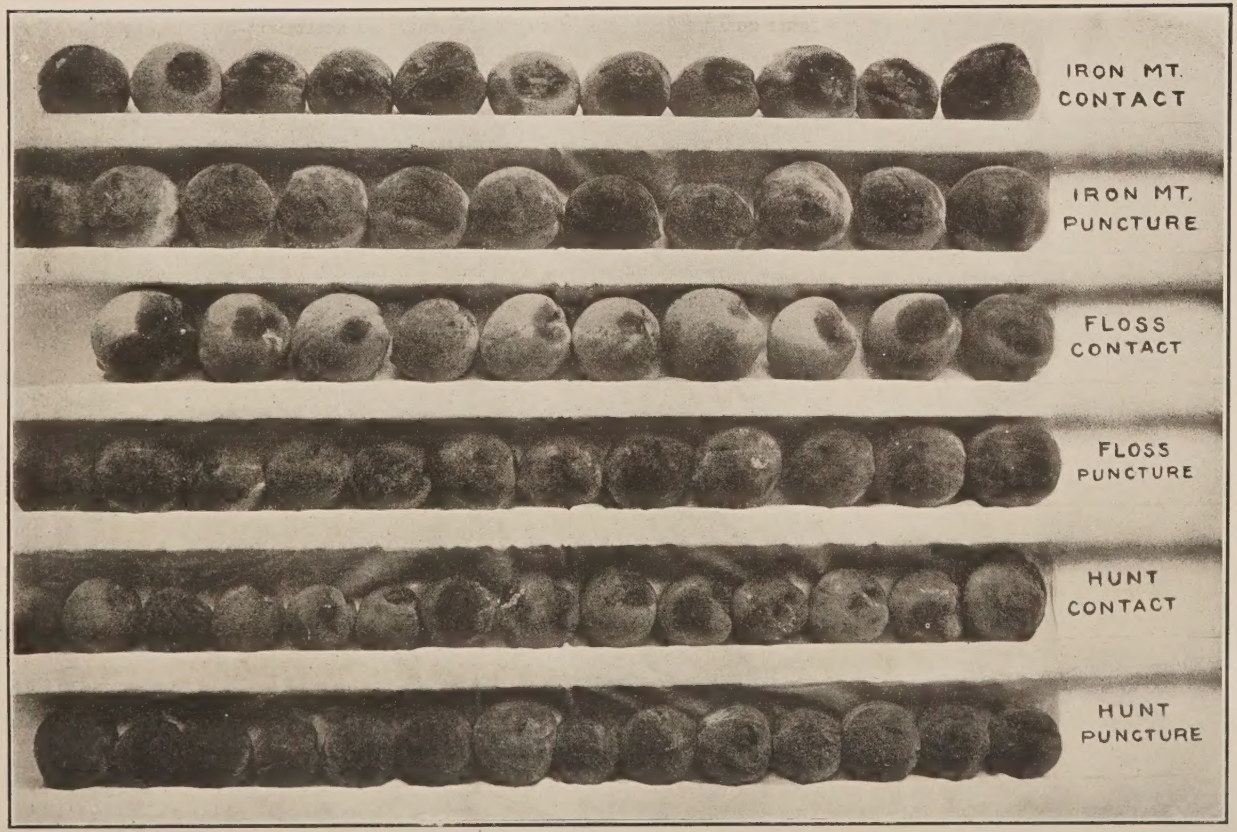

PLATE IV.—CONDITION OF PEACHES SHOWN IN PLATE III AFTER THEY HAD BEEN IN A WARM ROOM FOR TWO WEEKS.

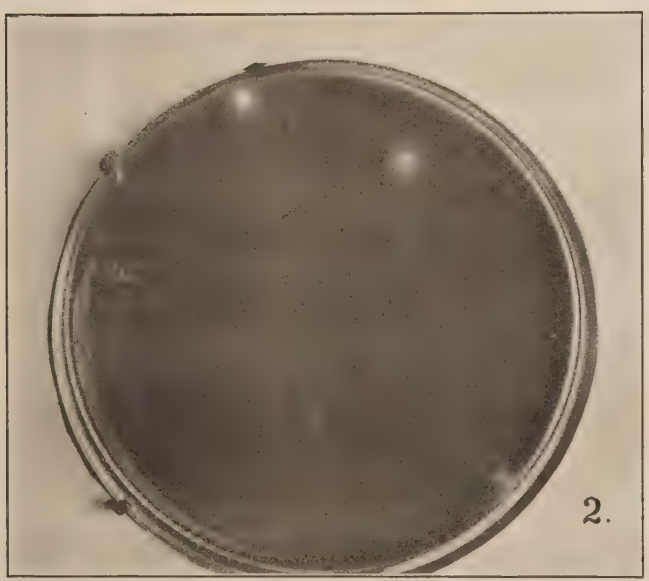

PLATE V.—PENICILLIUM GLAUCUM SPORES DESTROYED BY SULPHUR FUMIGATION. PETRI DISHES OF STERILE AGAR WERE EXPOSED ON THE FLOOR OF A ROOM ONE-HALF HOUR AFTER SPORES OF FUNGUS WERE DIFFUSED THROUGH THE AIR: 1, BEFORE FUMIGATION OF APPLES; 2, AFTER FUMIGATION OF APPLES.

FIG. 1

FIG. 2

PLATE VI.—APPLES INJURED BY SULPHUR FUMIGATION: 1, ESOPUS SPITZENBURG APPLE INJURED IN SOME UNKNOWN WAY (PROBABLY BY SULPHUR FUMIGATION); 2, ESOPUS SPITZENBURG APPLE INJURED BY SULPHUR FUMIGATION.

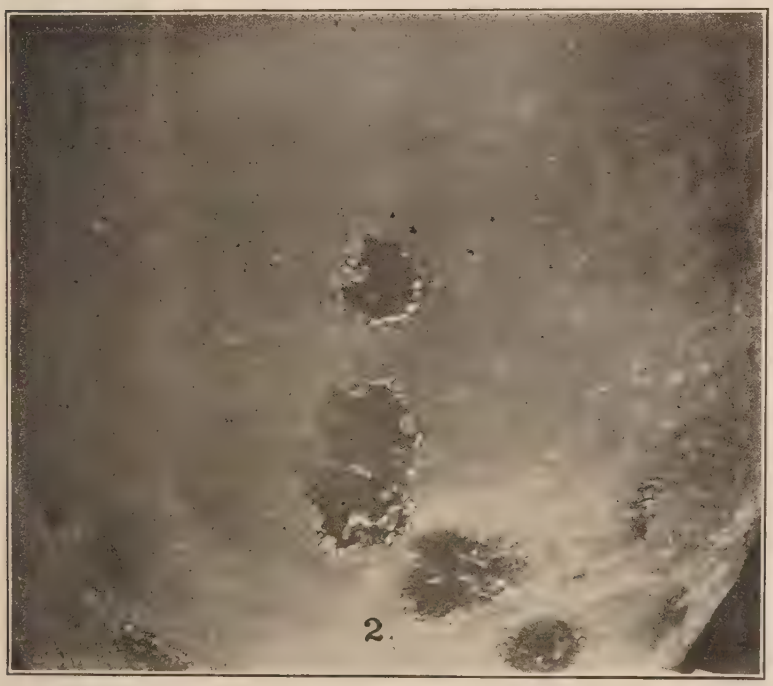

PLATE VII.—1, BOX OF ESOPUS SPITZENBURG APPLES INJURED IN SOME UNKNOWN WAY (PROBABLY BY SULPHUR FUMIGATION); 2, SCAB SPOT ON APPLE, MUCH ENLARGED, SHOWING THE MANNER OF GROWTH BENEATH THE EPIDERMIS.

TABLE II.—CONDITION OF INOCULATED APPLES REMOVED FROM COLD STORAGE TO WARM TEMPERATURE.

FIRST TEST.

Fungus.	Condition on May 16.	Condition on May 23.
Alternaria sp.	Decay has developed about the point of inoculation on all fruits.	Decay has continued to develop in every case.
Bitter rot, *Glomerella rufomaculans.*	Decay area about ½ inch in diameter has developed about the point of inoculation on all fruits.	Decay has continued to develop. Fungus is fruiting freely.
Black rot, *Sphæropsis malorum.*	Decay has developed to small extent.	Decay has affected about one-half of each fruit.
Blue mold, *Penicillium glaucum.*	Decay developed vigorously.	Decay spread over entire fruit.
Brown rot, *Sclerotinia fructigena.*	Decay developed vigorously in all varieties except Northern Spy, on which it has made small development, fungus fruiting most abundantly on Baldwin and R. I. *Grecning.*	Decay spread over entire fruit.
Pink rot, *Cephalothecium roseum.*	Decay has developed at the point of inoculation in each case.	Decay developed considerable fungus on some apples.

Plate II shows the condition of the apples on May 23.

The duplicate of this experiment was prepared on March 13, 1905. The same species of fungi and the same varieties of apples were used. The details of the inoculations were the same. A different cold storage house was used. A few hours after the inoculations were made in the laboratory the apples were stored in this house, being taken there by messenger. The temperature was recorded at different periods four times each twenty-four hours. An examination of these records shows that at 4 readings the temperature was 30°, at 3 it was

30½°, at 45 it was 31°, at 74 it was 31½°, at 90 it was 32°, at 25 it was 32½° and at 4 it was 33° F.

The apples and cultures were removed from the cold storage house on May 13, 1905 and at once brought to the laboratory by messenger and immediately examined. Their condition was as stated in the following table:

TABLE III.—CONDITION OF INOCULATED APPLES AND CULTURES AFTER BEING IN COLD STORAGE FOR TWO MONTHS.

SECOND TEST.

Fungus.	Growth in apples.	Growth in cultures.
Alternaria sp.	On some fruits there has been a slight growth, but nothing of importance.	No growth.
Blue mold, *Penicillium glaucum.*	Large decayed spots at the point of every inoculation.	Vigorous colonies in each culture.
Brown rot, *Sclerotinia fructigena.*	No growth in any of the fruits.	No growth.
Pink rot, *Cephalothecium roseum.*	No growth in any of the fruits.	No growth.
Scab, *Venturia inæqualis.*	No experiments with fruit.	Small growth in the cultures.

The condition of these apples and cultures upon removal from the cold storage house was the same as those in the first experiment. The experiments were comparable in every way and the results are practically identical.

As was done in the first experiment, the apples were photographed and then put away in a place at room temperature—about 70° during the day. They were examined in one week and again in two weeks, their condition being as follows:

TABLE IV.—CONDITION OF INOCULATED APPLES REMOVED FROM COLD STORAGE TO WARM TEMPERATURE.

SECOND TEST.

Fungus.	Condition on May 20.	Condition on May 27.
Alternaria sp.	Small decayed spots at the points of inoculation.	Decay has continued to develop.
Bitter rot, *Glomerella rufomaculans.*	Small decayed spots at point of every inoculation. Fungus fruiting on some.	Decayed spots have enlarged.
Black rot, *Sphæropsis malorum.*	Decay has developed at point of every inoculation.	Most of the fruits entirely decayed.
Blue mold, *Penicillium glaucum.*	Large part of every fruit has decayed.	Decay has spread.
Brown rot, *Sclerotinia fructigena.*	Decayed area of good size at nearly every point of inoculation.	Decay has spread.
Pink rot, *Cephalothecium roseum.*	Decay has started at the point of inoculation in most cases.	Decay has continued to develop.

These results agree in every respect with those of the first experiment and again demonstrate that the low temperature does not destroy the fungus, but simply retards its germination and growth.

THE EXPERIMENTS OF 1906.

The next year, the winter of 1906, another similar experiment was made. The same cold storage house was used as in the first experiment.

Pure cultures of the species of fungi used were obtained by making dilution plate cultures from material secured from natural infections. From these, sub-cultures on sugar beet plugs were made and the inoculations made from the last one after its pathogenicity had been tested.

Nine varieties of apples were used, some of each variety being inoculated with each species of fungus used. The usual precautions were taken to clean the fruit before the inoculations and to use sterile instruments in doing the work.

Immediately after the inoculations were made the fruit was taken by messenger to the cold storage house and placed at once in a room that contained other fruit where the temperature was 32° F.

The varieties used were Swaar, Fall Pippin, Twenty Ounce, Winter Banana, Deacon Jones, Fameuse, Reinette Pippin, Dickinson and Water. The same species of fungi were used as before, together with a few other species of no commercial importance.

The inoculations were made and the fruit placed in the cold storage house on January 18, 1906. It was removed, brought to the laboratory at Geneva, photographed and examined on March 22, thus being in the storage house 9 weeks.

The condition of the fruit upon its removal was as indicated in the following table:

TABLE V.—CONDITION OF INOCULATED APPLES AFTER BEING IN COMMERCIAL COLD STORAGE FOR NINE WEEKS.

THIRD TEST.

Fungus.	Growth in fruit.
Alternaria sp.	Decay has started at point of inoculation, but of very slight development.
Bitter rot, *Glomerella rufomaculans.*	Fruits all sound.
Black rot, *Sphæropsis malorum.*	Fruits all sound.
Blue mold, *Penicillium glaucum.*	Decay has started at all the points of inoculation, except in one fruit.
Brown rot, *Sclerotinia fructigena.*	Fruits all sound.
Pink rot, *Cephalothecium roseum.*	Fruits all sound.

These results agree with the previous experiments.

As was done in the other cases, the fruits were placed in a room where the temperature was about 60° F. during the day and somewhat less during the night.

At the end of one and two weeks the conditions were as follows:

TABLE VI.—CONDITION OF INOCULATED APPLES REMOVED FROM COLD STORAGE TO WARM TEMPERATURE.

THIRD TEST.

Fungus.	Condition of the fruit on March 31.	Condition of the fruit on April 7.
Alternaria sp.	Decayed spots enlarged in most of the fruits.	Decayed spots have continued to enlarge. Fungus is fruiting.
Bitter rot, *Glomerella rufomaculans.*	Slight decayed areas.	Decay developing and fungus fruiting at nearly every point of inoculation.
Black rot, *Sphæropsis malorum.*	Slight decayed areas.	Decay areas enlarging.
Blue mold, *Penicillium glaucum.*	Large decayed areas on all of the fruits.	All fruits completely decayed.
Brown rot, *Sclerotinia fructigena.*	Large decayed areas on most of the fruits.	Every fruit more than one-half decayed.
Pink rot, *Cephalothecium roseum.*	Fungus is growing, but decayed spots are small.	Fungus is growing and causing decay.

These results were also the same as in the previous experiments.

These experiments were made with the same species of fungus, using an assortment of varieties of apples, and the results are consistent in all cases. None of the fungi used proved to be capable of developing in the low temperature except the blue mold, *Penicillium glaucum*, which grew sparingly. The low temperature did not destroy any of the fungus spores, but simply retarded their germination. Upon removal to a higher temperature germination took place and decay of the fruit tissue resulted.

This inoculated fruit was in cold storage for two months or a little longer. It is possible, though not probable, that some of the species of fungi used would have developed and produced decay if the time had been extended.

All of the apples used in these experiments were kept in cold storage until shortly before being inoculated and hence did not go into the storage house in a warm condition. Had they been warm when they were placed in storage the kind of package used (bushel boxes) would have been quite important, for the fruit in a small package will cool down comparatively soon after going into a cool room. When large packages (barrels) are used, and the apples are warm, those in the center do not cool down to the temperature of the air of the room for some days.

Occasionally, barrels of apples are removed from cold storage and upon examination some of the fruit is found to be in a partly decayed condition, produced by the growth of some of the species of fungi that the preceding experiments have shown to be incapable of growing in the low temperature of a cold storage house. Assuming that the storage house was properly operated, such a condition of the fruit upon removal can be accounted for in two ways: (1) The apples may have been barreled and allowed to remain in the orchard or a shed or were on a railroad for some time. (2) It is possible also that decay would be found in barrels of apples that had been stored immediately after harvesting, if the weather at the time was warm. Under such conditions the fruit would go into the barrel warm and would be surrounded by warm air, and if the temperature of the fruit or air was 75° or 80° F. it would require about a week before the temperature of the fruit in the center of the barrel would be reduced to or near the temperature of the store room. But during the time the fruit was cooling decay could start and develop to some extent. Especially would this be true with a decay which develops quickly like bitter rot.

It is unfortunate that the only fungus that grows and produces decay in commercial cold storage is the most destructive and also most common species. But this rot is what might properly be termed a mechanical one, as the losses from it most often follow mechanical injuries to the fruit. Proper and careful handling will greatly reduce these injuries and therefore lessen the amount of decay from this rot.

AN EXPERIMENT IN TEMPERATURE OF 35° TO 56°.

An experiment similar to the previous one was made where the inoculated apples were placed in a temperature of from 35 to 56 F., the average being 47° F., determined by a maximum and minimum thermometer.

The same species of fungi were used as in the previous work and the varieties of apples were King, Baldwin, Rhode Island *Greening*, Northern Spy, Russet and Sutton.

The details of preparing the cultures, treating the apples and making the inoculations were the same as in the other experiments.

After the apples had been in this temperature for five weeks they were examined and all those inoculated with *Alternaria* sp., *Glomerella rufomaculans* (bitter rot) and *Sphæropsis malorum* (black rot) had developed decay spots ½ to 1 inch in diameter at the point of inoculation. The decayed spots in the apple inoculated with *Cephalothecium roseum* (pink rot) were smaller. Three-fourths of every apple inoculated with *Penicillium glaucum* (soft rot) was decayed. The culture of *Sclerotinia fructigena* (brown rot) which was used seemed to have lost its pathogenicity and the apples inoculated with this did not decay.

Petri dish cultures on potato agar were also made at the time the apples were inoculated and kept in the same room with the apples. In all of these cultures good growth of the different species of fungi developed.

At the end of the 5 weeks the apples were removed to a higher temperature and the decays developed rapidly.

AN EXPERIMENT IN A TEMPERATURE OF 48° TO 69°.

Another experiment was made, conducted in the same way as the previous ones, using a temperature of from 48° to 69° F., the average being 60.7° F., determined by a maximum and minimum thermometer.

At the end of 3 weeks the apples inoculated with *Alternaria* sp. and *Cephalothecium roseum* (pink rot) showed small decayed spots at the point of inoculation.

Those inoculated with *Sphæropsis malorum* (black rot), *Sclerotinia fructigena* (brown rot) and *Penicillium glaucum* (blue mold) were practically all decayed.

Those inoculated with *Glomerella rufomaculans* (bitter rot) showed decayed spots 1 to 2 inches in diameter at the point of each inoculation.

II. PEACH ROT IN COLD STORAGE.

The most severe and common decay of mature peaches is caused by *Sclerotinia fructigena* (brown rot). Since peaches are frequently put in commercial cold storage warehouses it was thought advisable to have some data regarding the behavior of this fungus in the temperature of the storage house.

Pure cultures of the fungus were secured by making dilution cultures with material from naturally infected fruit.

The varieties of peaches used were Iron Mountain, Floss and Hunt. A quantity of the fruit was secured and one-half of each variety inoculated by injecting some of the fungus from the pure culture beneath the skin. The other fruits were inoculated by contact which was done by first rolling them over paper moistened with distilled water and then over a quantity of the spores secured from several natural infections.

Immediately after the inoculations were made the fruits were taken by messenger to a commercial cold storage house in which the temperature was held at 32° F.

At the end of two weeks all of the fruits were removed and examined. Of those inoculated by puncture the conditions were as follows: Iron Mountain, 65 per ct. of fruits showed decayed spot ½ inch or less in diameter at the point of inoculation, 35 per ct. were entirely sound; Floss, 81 per ct. of the fruits showed decayed spot ¾ inch or less at point of inoculation, 19 per ct. were entirely sound; Hunt, 56 per ct. showed decayed spot 1½ inch or less at point of inoculation, 44 per ct. were entirely sound. The condition of some average specimens of these fruits is shown in Plate III.

None of the fruits inoculated by contact showed any decay at the end of two weeks.

When the fruits were taken out of cold storage they were kept in a room where the temperature was rather low, so the decay did not develop rapidly. But at the end of ten days the fruits inoculated by puncture were all decayed. Those inoculated by contact were as follows: Iron Mountain, all of the fruits decayed; Floss, 71 per ct. decayed, 29 per ct. sound; Hunt, 80 per ct. decayed, 20 per ct. sound. The condition of some of these fruits is shown in Plate IV.

These results indicate that the development of the brown rot in peaches is practically checked while the fruit is in commercial cold storage, even though the fungus can enter the fruit through a break in the epidermis. Peaches with a sound epidermis rolled in spores of the brown rot fungus did not develop the decay while in cold storage.

III. SULPHUR FUMIGATION TO DESTROY APPLE ROT FUNGI.

Experiments have been made to determine if it is possible by fumigation to destroy the spores of the various species of fungi that cause the decay of apples. If this were practicable it might be desirable to do so at the time the fruit storage house is being cleaned in preparation for the new crop. Especially would this be the case in an ordinary storage room where fruit is often allowed to decay and remain for some time.

The method of conducting these experiments was to first inoculate a quantity of apples with pure cultures of the various species of fungi. When the decay had developed in these fruits and the fungus was fruiting freely, cultures would be made by transferring some of the fungus to a sterile sugar beet plug, thus determining whether it was in an active condition. The fruits would then be placed in a tight room and different amounts of sulphur burned. Upon opening the room, usually a day later, cultures would again be made from the apples, thus determining whether it was active or had been destroyed by the fumigation. Experiments were also made by blowing the spores of *Penicillium glaucum* (blue mold) in the air and then fumigating and determining if this destroyed the spores.

The apples used had been artificially inoculated with the following species of fungi which were fruiting freely on decayed areas: *Glomerella rufomaculans* (bitter rot), *Sphæropsis malorum* (black rot), *Penicillium glaucum* (blue mold), *Sclerotinia fructigena* (brown rot), *Cephalothecium roseum* (pink rot) and *Rhizopus* sp.

Before the fumigation, cultures were made of these fungi by transferring some of the spores to sterile sugar beet plugs in test tubes. When this was done the apples were placed in a tight room 8x9x10 feet equal to 720 cubic feet. Four ounces of flowers of sulphur was placed in a dish and burned.

The room was kept closed for about 24 hours. When opened, cultures were again made in the same way as before, thus determining whether the spores had been destroyed by the fumigation.

The results indicated that all but the spores of *Penicillium glaucum* (blue mold) were destroyed. These were growing vigorously.

Repetitions of this experiment were made, using ½, 1, 1½ and 2 pounds of sulphur. One and one-half pounds of sulphur burned in a tight room which was kept tightly closed for about 24 hours gave fairly satisfactory results, but 2 pounds of sulphur gave entirely satisfactory results.

In this experiment there were used a large number of apples, which had been artificially inoculated and were producing an abundance of spores of the fungus.

After transfer cultures were made from all of these apples they were placed in a room which was made as tight as possible and the fumigation was done as follows: A large pan placed on the floor in the center of the room was filled with a quart or more of water, two bricks were set edgewise in this pan and on them rested a small tin pan which contained the 2 pounds of sulphur. This was easily ignited in the place where a teaspoonful of alcohol had been poured. The arrangement of the sulphur over water was done to eliminate any danger there might be from fire.

The room was closed immediately after the sulphur was ignited. About 24 hours later it was opened. Cultures were

again made from all the apples. After a reasonable time was allowed for their growth they were all examined. Those made before fumigation were growing vigorously while those made after the fumigation had not grown at all. They were all kept under observation for some time, but no growth developed in any cultures made after fumigation.

The experiments to destroy the spores of *Penicillium glaucum* (blue mold) in the air were made by fumigating with 1 and 1½ pounds of sulphur to 720 cubic feet of space. The results with 1 pound indicated that it was insufficient, but 1½ pounds was used with good results.

The method was to secure a quantity of apples that were well covered with the spores of *Penicillium glaucum* (blue mold) part of them being placed on shelves in the room where the fumigation was done, the others set on a plate and by vigorous fanning the spores were blown all about the room. One-half hour later 12 petri dishes containing sterile potato agar were placed on the floor of the room and their covers removed for exactly 3 minutes. During this time the operator remained in the room so as not to cause any unnecessary air currents by opening the door to go out.

The fumigation was then done in the way previously described. The room was kept closed for about 24 hours. When opened the apples that were left in were set on a plate and the spores on them blown about the room. One-half hour later 12 petri dishes of sterile potato agar were exposed for 3 minutes in the same way as was done before fumigation.

A few days later an examination was made of all the plates. Those exposed before fumigation were completely filled with colonies of the blue mold. In the 12 plates exposed after fumigation most of them were entirely sterile and in the others there were only a few scattered colonies, which very evidently came as a result of contamination. See Plate V.

The results of these experiments indicate that the spores of the fungi that cause the most common decays of apples, whether on the decayed fruit or floating in the air, can be destroyed by sulphur fumes, using about 1 ounce of sulphur to 25 cubic feet of space.

IV. APPLE INJURY BY SULPHUR FUMIGATION.

In March, 1905, a box of injured Esopus Spitzenburg apples was received at this Station with a request to diagnose the trouble, if possible. The apples were grown and packed in the State of Washington and shipped to New York where they were placed in cold storage. Upon being removed, some of the boxes of fruit showed the trouble while others from the same car were sound.

The fruit was of the first grade and each apple wrapped in paper. The financial loss was important, as a considerable amount of high priced fruit had been ruined from a commercial standpoint.

Scattered irregularly over the surface of each apple were conspicuous spots of various sizes where the epidermis was dead, discolored and slightly sunken. Each spot was nearly circular, though on some apples the adjacent spots had coalesced, forming a large affected area of irregular shape. Beneath each spot to the depth of a few millimeters the flesh was dead, shrunken and dry, appearing as though affected with a dry rot. See Plate VI, fig. 1. There was no disagreeable odor or taste to the dead flesh or epidermis.

In the center of each of the smaller spots, and scattered over the larger affected areas, were small bodies resembling the pycnidia of a fungus, but examination showed them to be only the normal lenticels of the apples. Failure to find either fungi or bacteria as a cause of the injury led to the belief that some treatment of the fruit, such as fumigation, might be the cause. Sulphur, being commonly used for fumigation, was experimented with to note the effect of the fumes upon ripe apples. Fruits of different varieties, including Esopus Spitzenburg were placed in a bell jar which was then filled with sulphur fumes. After five minutes the fruit was removed and found to have developed numerous spots that were in every way identical with those on the apples received for examination. Plate VI, fig. 2.

The experiment was repeated many times with wet and dry fruits, but the characteristic spots were always produced,

though more conspicuous on red apples. The spots continued to enlarge for some time after the fruits were removed from the fumes.

The presence of a lenticel in the center of each spot would indicate that the sulphur dioxid passes into the fruit at this point and causes the bleaching of the tissue. A similar effect was produced when an artificial break in the epidermis was made. A lenticel makes a strong color contrast with the bleached epidermis, thus giving it the appearance of a pycnidium. Hydrocyanic acid gas was also tried but it did not injure the fruits, though a strong application was made for a good length of time.

These were the only two substances used, it is possible that other chemicals would produce a similar injury.

Why there should ever have been a desire to fumigate the fruit with anything is not clear. Where it was done and by whom could not be determined. It was first class fruit in every respect and certainly could not have been improved by any known treatment. It is possible that it was done by mistake when a car or store house was being fumigated.

The incident is valuable in bringing out the danger of fumigating fruit with sulphur, even though these apples may have been injured in some other way.

V. ENLARGEMENT OF APPLE SCAB SPOTS UNDER COVERING OF BORDEAUX MIXTURE.

The question often arises, will the development of a scab spot, *Venturia inæqualis,* upon an apple be checked if it is thoroughly covered with bordeaux mixture? Opinions upon this subject seem to have differed. Fairchild[1] states that the scab spot will grow beneath a layer of bordeaux mixture. Craig[2] states "that it is true that a scab spot will cease to enlarge if thoroughly covered with bordeaux mixture" and

[1] Fairchild, D. G. Bordeaux mixture as a fungicide. U. S. Dept. of Agr., Div. of V. P., Bul. 6, p. 43.

[2] Craig, John, and Van Hook, J. M. Pink rot an attendant of apple scab. Cornell Sta. Bul. 209, p. 167.

Warren[3] reports that the spot will continue to grow though covered with spray.

These reports appear to have been based upon general observations. Growers frequently want to know definitely about the matter as it has an important bearing upon the advisability of a late summer spraying in seasons when scab is especially severe. That information based upon experimental data might be secured, the following tests were undertaken:

On August 1, a number of apples showing well defined scab spots were found upon a Carolina June tree. They were conspicuously marked and the scab spot or spots to be under observation were numbered by marking upon the fruit with water proof ink and then making careful measurements of the spots. Part of the apples were thoroughly sprayed with bordeaux mixture, the others being reserved for checks. The bordeaux was the 5-5-50 formula and applied very thoroughly especially on the scab spots.

Frequent examinations were made of these apples and measurements and notes taken. In all but one case the spots that were covered with the bordeaux mixture continued to enlarge, as did all of the spots that were marked for checks.

Similar tests were made on August 3 with Rhode Island *Greening* and Fall Pippin. On August 6 with the same varieties and on August 30 with Rhode Island *Greenings.* In every one of these tests all of the scab spots enlarged after they had been covered with a thick coating of bordeaux mixture.

These results are what would be expected, as it is known that the scab fungus grows in the fruit beneath a thin outer layer known as the epidermis. See Plate VII, fig. 2. The margins or growing portion of the scab spot is then protected and beyond the reach of the action of the bordeaux mixture.

[3] Warren, G. F. An apple orchard survey of Wayne County, New York. Cornell Sta. Bul. 226, p. 339.